PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 344472D19980	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/IB 03/02076	International filing date *(day/month/year)* 18.04.2003	Priority date *(day/month/year)* 19.04.2002
International Patent Classification (IPC) or both national classification and IPC C07K14/705, C07K14/705		
Applicant CENTRE NATIONAL DE LA RECHERCHE ... et al.		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 7 sheets, including this cover sheet.

   ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 3 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the opinion
   - II ☐ Priority
   - III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
13.11.2003	04.06.2004
Name and mailing address of the international preliminary examining authority: European Patent Office - P.B. 5818 Patentlaan 2 NL-2280 HV Rijswijk - Pays Bas Tel. +31 70 340 - 2040 Tx: 31 651 epo nl Fax: +31 70 340 - 3016	Authorized Officer Schmidt, Harald Telephone No. +31 70 340-4023

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/IB 03/02076

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:

Description, Pages

1-28 as originally filed

Claims, Numbers

1-23 filed with telefax on 13.04.2004

Drawings, Sheets

1/7-7/7 as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

   These elements were available or furnished to this Authority in the following language: , which is:

   ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).

   ☐ the language of publication of the international application (under Rule 48.3(b)).

   ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

   ☒ contained in the international application in written form.

   ☒ filed together with the international application in computer readable form.

   ☐ furnished subsequently to this Authority in written form.

   ☐ furnished subsequently to this Authority in computer readable form.

   ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

   ☒ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

   ☐ the description, pages:

   ☒ the claims, Nos.: 24

   ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been examined in respect of:

   ☐ the entire international application,

   ☒ claims Nos. 17 (as to IA)

   because:

   ☒ the said international application, or the said claims Nos. 17 (as to IA) relate to the following subject matter which does not require an international preliminary examination (specify):

   see separate sheet

   ☐ the description, claims or drawings *(indicate particular elements below)* or said claims Nos. are so unclear that no meaningful opinion could be formed *(specify)*:

   ☐ the claims, or said claims Nos. are so inadequately supported by the description that no meaningful opinion could be formed.

   ☐ no international search report has been established for the said claims Nos.

2. A meaningful international preliminary examination cannot be carried out due to the failure of the nucleotide and/or amino acid sequence listing to comply with the standard provided for in Annex C of the Administrative Instructions:

   ☐ the written form has not been furnished or does not comply with the Standard.

   ☐ the computer readable form has not been furnished or does not comply with the Standard.

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-23
	No:	Claims	
Inventive step (IS)	Yes:	Claims	1-23
	No:	Claims	
Industrial applicability (IA)	Yes:	Claims	1-16,18-23
	No:	Claims	

2. Citations and explanations

see separate sheet

Re Item III

Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

Claim 17 relates to subject-matter considered by this Authority to be covered by the provisions of Rule 67.1(iv) PCT. Consequently, no opinion will be formulated with respect to the industrial applicability of the subject-matter of the claim (Article 34(4)(a)(I) PCT).

Re Item V

Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

The following documents (D) are referred to in this communication:

D1: Finckh U et al. (2000) Am.J.Med.Gen. 92: 40-46 & EBI-Database (1999), Accession number Q9XT41
D2: Castellani V et al. (2000) Neuron 27: 237-249
D3: WO 99/04263 (28.01.1999)

Novelty

Document D1 discloses the amino acid sequence of neuronal cell adhesion molecule L1 (L1CAM) comprising the sequence FASNKLGTAMS (aa 110-120). It is further stated that L1CAM is involved in a wide range of neuronal regulation systems (see page 40). Document D2 discloses that L1Fc chimeric molecules converts Sema3A-induced repulsion into attraction (see page 242, right-hand column).

However, both documents are silent over peptides having no more than 10 amino acids.
Therefore, subject-matter of claims 1 to 23 is novel in the sense of Article 33(2) PCT.

Inventive step

The document D2 is considered to represent the most relevant prior art for the present application and discloses a multiprotein complex wherein NP-1 binds to L1, plexin and Sema-3A. By binding to the complex, L1 regulates the cytoplasmic cGMP level. It is further stated that L1 associates with several other proteins, e.g. tyrosine kinases (see pages 246 and 247).
D2 also teaches that soluble L1 blocks growth cone collapse in response to Sema3A, a process that is well known to be accompanied by endocytosis (see figure 6).

Subject-matter of claims 1 to 5,17 and 21 to 23 differs in that distinct peptides derived from L1 are used.

Subject-matter of claims 6 to 16 and 18 to 20 differs in that methods for the identification of further binding partners are provided.

The problem to be solved by the current application is the provision and identification of further compounds for the regeneration of neuronal/axonal regeneration or for treating neurodegenerative diseases.

Since there is no suggestion in the prior art cited to use the compounds of claims 1 to 5 to solve the problem posed, subject-matter of claims 1 to 5, 17 and 21 to 23 involves an inventive step in the sense of Article 33(3) PCT.

To provide a method for identifying compounds that bind the NP-1 protein and alter the binding of NP-1 and L1 and/or block Sema3A-induced endocytosis is not regarded to be obvious for the skilled person, since D2 does not disclose or suggest methods of identifying such compounds and D3 wherein methods are taught to identify compounds that bind to neuropilins for the treatment of neurodegenerative diseases does not mention the transmembrane glycoprotein L1.
Therefore, subject-matter of claims 6 to 16 and 18 to 20 involves an inventive step in the sense of Article 33(3) PCT.

Industrial applicability

For the assessment of the present claim 17 on the question whether it is industrially applicable, no unified criteria exist in the PCT Contracting States. The patentability can

also be dependent upon the formulation of the claims. The EPO, for example, does not recognize as industrially applicable the subject-matter of claims to the use of a compound in medical treatment, but may allow, however, claims to a known compound for first use in medical treatment and the use of such a compound for the manufacture of a medicament for a new medical treatment.

Claims

1. A peptide comprising SEQ ID N° 1 and no more than 10 amino acids preferably, inducing attraction of the axonal growth, in particular in presence of the semaphorin (Sema3A, L1 and NP-1 proteins.

2. The peptide of claim 1 consisting of SEQ ID N° 1.

3. The peptide of claim 1, comprising or consisting of SEQ ID N° 2.

4. The peptide of claim 1, consisting of SEQ ID N° 3.

5. An inducer of axonal growth consisting of the peptide of anyone of claims 1 to 4.

6. A method for identifying a compound able to bind the NP-1 protein, comprising the steps of:

a) contacting said compound with a sample comprising the NP-1 protein, before, after or concomitantly with contacting said sample with the L1-Fc protein or the peptide of any of claims 1 to 4,

b) studying the binding between NP-1 and L1-Fc or said peptide,

the binding of said compound to the NP-1 protein being deduced from the alteration of the binding of NP-1 with L1-Fc or said peptide observed in absence of said compound.

7. The method of claim 6, wherein said sample also comprises the L1 protein.

8. The method of claim 6 or 7, wherein said NP-1 protein is a recombinant protein.

9. The method of anyone of claims 6 to 8, wherein said sample comprises a cell expressing the NP-1 protein at its surface.

10. The method of any of claims 6 to 9, wherein said sample comprises a cell expressing the NP-1 and L1 proteins at its surface.

11. The method of claim any of claims 6 to 10, wherein said L1-Fc protein or said peptide is labeled and said binding of NP-1 protein with said L1-Fc protein or said peptide is assayed by level of signal associated with said NP-1 protein.

12. The method of any of claims 6 to 11, wherein said alteration of binding of NP-1 with L1-Fc or said peptide is assayed by measuring a secondary signal appearing in the presence of said binding.

13. The method of claim 12, wherein said secondary signal is induced in the presence of Sema3A.

14. The method of claim 13, wherein said secondary signal is the reversion of the repulsory effect of Sema3A on axonal growth.

15. The method of claim 13, wherein said secondary signal is the activation of NO synthesis.

16. The method of claim 13, wherein said secondary signal is an increased production of cGMP.

17. A method for attracting /modulating the direction of axonal growth by a cell suited to promote axonal growth, submitted to a Sema3A flow, comprising the step of contacting said cell to the peptide of any of claims 1 to 4, or the inducer of claim 5.

18. A method for identifying a compound able to revert the repulsory effect of Sema3A on the axonal growth from a suited cell, comprising the steps of contacting said cell expressing L1 and NP-1 at its surface with said compound in the presence of Sema3A, and studying the increase in NO synthesis in said cell.

19. A method to screen for molecules that prevent the internalization of the dextran induced by the Sema3A treatment or to screen for molecules capable of blocking Sema3A- induced endocytosis by cells, characterized in that said method comprises the following steps of:

a) culturing cells expressing L1 and NP-1 in the presence of Sema3A, a labelled dextran and the molecule to be assayed ; and

b) visualizing or determining if said labelled dextran has been internalized into the cells ;

c) selecting the assayed molecule if no uptake of the labelled dextran by the cells could be detected.

20. A method to screen for molecules capable of blocking Sema3A- induced co-internalization of L1 and NP-1 proteins, to screen molecules capable of maintaining cell surface expression of L1 and NP-1 proteins or to screen molecules capable of blocking

Sema3A- induced endocytosis, characterized in that said method comprises the following steps of:

a) culturing cells expressing L1 and NP-1 in the presence of Sema3A and the molecule to be assayed ; and

b) visualizing or determining if said L1 and NP-1 proteins have been co-internalized into the cells using an immunocytochemical method in presence of antibodies directed against L1 and NP-1 proteins ;

c) selecting the assayed molecule if L1 and NP-1 proteins could be detected at the surface of the cells.

21. Use of the peptide of anyone of claims 1 to 4, or the inducer of claim 5, for the manufacture of a drug for neuronal/axonal regeneration.

22. Use of the peptide of anyone of claims 1 to 4, or the inducer of claim 5, for the manufacture of a drug for treating neurodegenerative diseases.

23. Use of the peptide of anyone of claims 1 to 4, or the inducer of claim 5, as a targeting agent, in the manufacture of a composition to be used in cell therapy.